Docket No. 1662/55602

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Inventor	:	LIDOR-HADAS et al.
Application No.	:	10/045,970
Filed	:	January 11, 2002
For	:	A NOVEL PROCESS FOR PREPARING PURE ONDANSETRON HYDROCHLORIDE DIHYDRATE
Examiner	:	Laura Lynne Stockton
Art Unit	:	1626

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Alexandria, VA 22313-1450

DECLARATION UNDER 37 C.F.R. § 1.132

Sir:

I, Dr. Ramy Lidor-Hadas, hereby declare and state as follows:

1. I am a co-inventor, with Eliezer Bachar, of the subject matter disclosed and claimed in the above-identified patent application. I am experienced in the field of pharmaceuticals, and my *Curriculum Vitae* is attached as Exhibit A.

2. Ondansetron hydrochloride dihydrate, as described in Chen (Zhongguo Yiyao Gongye Zazhi (1993), 24(6), pages 241-242), Tyers (U.S. Pat. No. 4,845,115) ("Tyers 1"), Coates (U.S. Pat. No. 4,695,578), and Tyers (U.S. Pat. No. 4,835,173) ("Tyers 2"), does not have a purity of at least about 99.0%.

3. The Coates process (Example 7 followed by Example 10) yields crude ondansetron hydrochloride dihydrate containing 0.4% exo-methylene. The Coates purification process yields ondansetron hydrochloride dihydrate containing 0.12% exo-methylene. In contrast, the purified ondansetron hydrochloride dihydrate of the present invention has only 0.01% exo-methylene.

4. The purity and color of ondansetron hydrochloride dihydrate is greatly improved by using water and activated carbon rather than water/isopropanol. The

attached Exhibit B, at pages 4-7, shows supporting experimental data. Even when starting with ondansetron base containing greater than 1% exo-methylene, crystallization with activated carbon gave ondansetron hydrochloride dihydrate with a purity of at least about 99.0%.

5. The purity of ondansetron hydrochloride dihydrate is also greatly improved by using about 4 to about 6 equivalents methyl-imidazole in the preparation of the ondansetron base starting material. The attached Exhibit B, at pages 1-3, shows that exo-methylene formation is inversely related to the amount of methyl-imidazole used. The Coates process uses 3 eq. methyl-imidazole.

6. I further declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that the undersigned acknowledges that any false statements and the like so made are punishable by fine or imprisonment or both under §1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of any patent that issues from U.S. Application Serial No. 10/045,970.

Date: 25/July/2004.

Ramy Lidor-Hadas

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CURRICULUM VITAE

15-5-2003

Born: 24. Oct. 1951

Place: Tel-Aviv

Married +2

1998-Now	TEVA Ltd., Manager of the process chemistry department, R&D division.
1991-1998	TEVA Ltd., Corporate R&D, process development team leader.
1986-1991	ABIC Ltd., R&D, Senior chemist.
1985-1986	Bromine Compounds Ltd., in collaboration with Prof. S. Rozen, school of chemistry, Tel-Aviv university.
1984- 1985	Research Associate with Prof. Y. Kashman, school of chemistry, Tel-Aviv university.
1984	Post doctoral research, Prof. P. A. Bartlett, U. C. Berkeley, California. USA.
1978-1984	Tel-Aviv university, Ph. D. Thesis, "Synthetic uses of oxime ethers" under the supervision of Prof. S. Shatzmiller.
1976-1978	Tel-Aviv university, M. Sc. thesis "Tetrabromocyclopentadienone dimer" under the supervision of Prof. B. -Z. Fuchs
1973-1976	Tel-Aviv university, B. Sc. (Magna cum lauda).

Presentation in conferences

1996 Israel chemical society meeting (Poster).

1983 The 4th Israel-Germany Güntner symposium of chemistry (9-14, Oct.).

1982 Israel chemical society meeting.

Awards

1983 M. Landau award for research in chemistry (8 Nov).

Grants

1984 Rotchield Foundation for post doctoral research.

Advanced study and courses

2000 Developments in industrial crystallization (Continuing education and training, University of Manchester institute of science and technology (UMIST), 29th-31st March 2000).

1999 Basic principles of chemical engineering (The center for professional advancement, Amsterdam, 28th June-2nd July 1999).

1998 Synthesis and methods (Scientific Update, Florence, 6-10th April 1998).

1995 Industrial synthesis of optically active compounds (Scientific Update, Florida, 6-8th November 1995).

1994 Introduction to industrial pharmacy (Teva and the Hebrew university, January-March 1994, 84 hours).

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Articles

1. **Bromo-organics. 1. Tetrabromocyclopentadienone-dimer: unusually great reactivity to nucleophilic attack on bridged carbonyl.**
   Fuchs, B., Lidor, R., Krüger, C. & Liu L. -K., *Nouv. J. Chim.*, 4(6), 361-3 (1980).

2. **Bromo-organics. 2. Irradiation-induced transformations of tetrabromocyclopentadienone dimer**.
   Fuchs, B., Drucker, C. & Lidor, R., *J. Org. Chem.*, **46**(7), 1479-81 (1981).

3. **Highly regiocontrolled and rapid lithiation of 3-methyl-4H-5,6-dihydro-1,2-oxazine: Elaboration for α-methylene ketone synthesis.**
   Lidor, R. & Shatzmiller, S., *J. Am. Chem. Soc.*, **103**(19), 5916-17 (1981).

4. **(2-Chloralkylidene)amine-oxides as alkylation reagents; Reaction with amines and the anions of dialkyl malonates.**
   Lidor, R. & Shatzmiller, S., *Liebigs Ann. Chem* (2), 226-30 (1982).

5. **Synthesis and reactions of 3-isoxazolines.**
   Shatzmiller, S., Shalom, E., Lidor, R., & Tartkovski, E., *Liebigs Ann. Chem* (6), 906-12

(1983).

6. **Geometrically controlled dimerization of acetone O-methyloxime; New way to synthesize pyrrole derivatives.**
Shatzmiller, S. & Lidor, R., *Synthesis*, (7), 590-93 (1983).

7. **A novel route to arylacetones *via* a masked α-acylcarbonium intermediate.**
Shatzmiller, S., Lidor, R., Shalom, E. & Bahar, E., *J. Chem. Soc. Chem. Commun.*,(12), 795-6 (1984).

8. **Latrunculins: NMR study, two new toxins and a synthetic approach.**
Kashman, Y., Groweiss, A., Lidor, R., Blasberger, D. & Carmely, S., *Tetrahedron,* **41**(10), 1905-14 (1985).

9. **Synthetic studies related to latrunculin. Synthesis of tetrahydropyranylthiazolidin-2-one systems.**
Kashman, Y., Lidor, R., Blasberger, D. & Carmely, S., *Tetrahedron Let.*, **27**(12), 1367-70 (1986).

10. **Regiocontrolled synthesis of 4-halo-5,6-dihydro-4H-1,2-oxazines; A novel route for α-fluorovinyl ketones.**
Shatzmiller, S., Lidor, R. & Shalom, E., *Isr. J. Chem.*, **27**(1), 33-8 (1986).

11. **Aromatic bromination using bromine fluoride with no Friedel-Crafts catalyst.**
Rozen. S., Brand, M. & Lidor, R., *J. Org. Chem.*, **53**(23), 5545-7 (1988).

12. **Geometry-directed generation and synthetic application of α-lithio oxime ethers.**
Shatzmiller, S., Lidor, R., Shalom, E., Menashe, N. & Bahar, E., *Isr. J. Chem.*, **29**(2-3), 187-94 (1989).

13. **Reaction of α-lithio O-alkyloximes with diiodomethane - a synthesis of α-iodomethylene oxime ethers.**
Shatzmiller, S., Lidor, R. & Bahar, E., *Liebigs Ann. Chem* (4), 381-3 (1991).

14. **Reactions of lithium salts of O-alkyoximes with halogens: A highly stereoselective oxidative coupling of 5,6-dihydro-3-methyl-4H-1,2-oxazine.**
Shatzmiller, S., Lidor, R., Bahar, E. & Goldberg, I., *Liebigs Ann. Chem* (9), 851-6 (1991).

15. **Lithium enamindes - lithium salts of azomethine derivatives.**
Shatzmiller, S. & Lidor, R., Chapter 25 in "*The chemistry of enamines*", Edited by Z. Rappoport, John Wiley & Sons, 1994.

16. **A facile synthesis for racemic and optically active 1-aminoindans.**
Lidor, R., Bahar, E., Zairi, O., Atili, G. & Amster, D., *Organic preparations and procedures international*, **29**(6), 701-706 (1997).

17. **Latent fingerprint visualization by 1,2-indanedione and related compounds: preliminary results.**
Almog, J., Springer, E., Wiesner, S., Frank, A.,Khodzhaev, O., Lidor, R., Bahar, E., Varkony, H., Dayan, S.& Rozen, S., J. Forensic Sci., **44**(1), 114-118 (1999).

18. **Reagents for the chemical development of latent fingerprints: Scope and limitations of benzo[f]ninhydrin.**
Almog, J., Springer, E., Wiesner, S., Lidor, R., Bahar, E., J. Forensic Sci., (to be published in May 2000).

Patents

1. **Process for the ring bromination of benzenoid compounds.**
Rozen, S., Lidor, R. & Brand, M., Israeli Patent No. 0079627 A1, Aug. 5, 1986, Bromine compounds Ltd.

2. **Optical resolution of threo-2-hydroxy-3-(2-aminophenylthio)-3-(4-methoxyphenyl) - propionic acid.**
Lidor. R. & Singer, C., US Patent, 5,302,741. Apr. 12, 1994, Teva Pharmaceutical. Industries Ltd.

3. **Method for preparing optically active aminoindan derivatives.**
Lidor R. & Bahar E., US Patent 5,639,913, Jun. 17, 1997, Teva Pharmaceutical. Industries Ltd.

4 **Process for preparing alendronic acid**
Lidor-Hadas R. & Lifshitz R., US Patent 6,201,148 B1, Mar. 13, 2001, Teva Pharmaceutical. Industries Ltd.

5. **Process for manufacturing of L-dopa ethyl ester**

   Lidor R., Bahar E. & Frenkel A., US Patent 6,218,566 B1, Apr. 17, 2001, Teva Pharmaceutical. Industries Ltd.

6. **Hydrate forms of Alendronate sodium, processes for manufacture thereof, and pharmaceutical composition thereof**

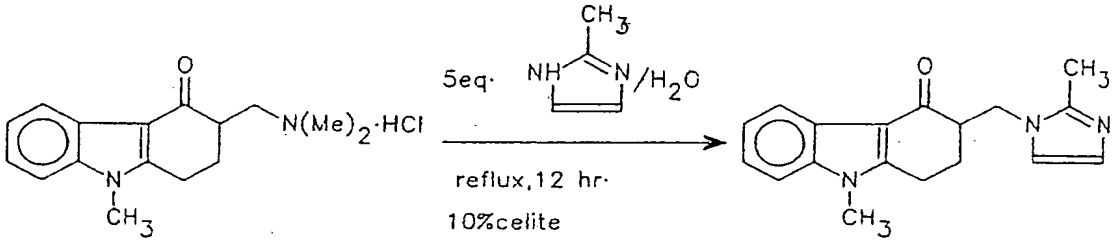
   Finkelstein N., Lidor-Hadas R. & Aronheim J., US Patent 6,281,381 B1, Aug. 28, 2001, Teva Pharmaceutical. Industries Ltd.

7. **Hydrolysis of Atorvastatin ester derivatives with calcium hydroxide to give Atorvastatin hemi-calcium salt.**

   Niddam V., Lidor-Hadas R., Lifshitz R. & Ishai E., US Patent 6,528,661 B2, Mar. 4, 2003, Teva Pharmaceutical. Industries Ltd.

8-21 **Patent submission (PCT and provisional)**

ONDANSETRON BASE (OND-BASE)

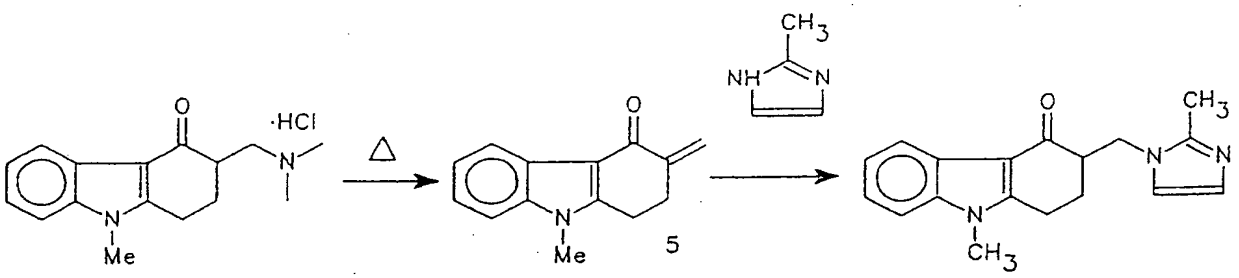


dimethylamino–Me–carbazolone
(DMA–Me–CAR)

ondansetron base[19]
(OND–base)

a) Starting materials, solvent, reaction time and mechanism

This stage required the hydrochloride salt of DMA-Me-CAR as starting material rather than the base. It was possible to start with the free base which was converted in situ to the HCl salt by aqueous HCl (EB-292). The first step of the reaction is probably the Hofmann elimination to form exo-methylene intermediate which further reacts with imidazole to yield OND-base:

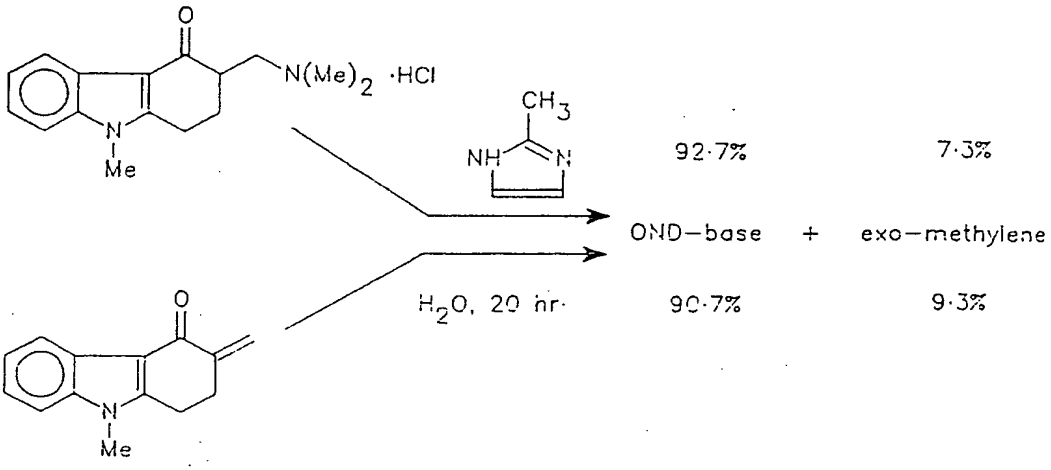


DMA–Me–CAR
hydrochloride

exo–methylene–CAR

OND–base

Support for such a mechanism was the fact that similar product mixtures were obtained when OND-base was synthesized either from DMA-Me-CAR or from exo-methylene:



In the original patent (reference 1b, example 7, page 19) the reaction was performed in 10 volumes of refluxing water for 20 hours. We used the same solvent and found that 12 (EB-306) or 20 (EB-308) hours gave the same results.

b) Equivalents of 2-methylimidazole (MI) and work-up procedure

There was an indirect relationship between the amount of MI used in this reaction and the exo-methylene by-product formation. We employed 3,4 and 5 equivalents of MI and examined the HPLC area % of the exo-methylene at OND.base, both crude and crystallized. The results are summarized in Table 1.

TABLE 1

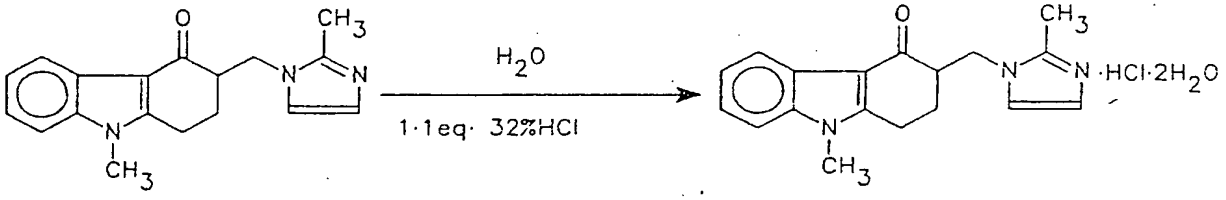
REFERENCE		EB-308		OZ-371		AD-283	
MI EQUIVALENTS		3		4		5	
OND BASE		CRUDE	CRYST	CRUDE	CRYST	CRUDE	CRYST
HPLC AREA %	EXO-METHYLENE	7.3	1.47	5.6	0.96	4.8	0.74
	OND BASE	90.0	98.1	91.7	98.3	92.9	98.6
	DMA-Me-CAR	1.40	0.23	1.80	0.40	1.60	0.40

Even though we developed a method to reduce the by-product level to less than 0.1% starting from levels as high as 2.5%, we preferred to use 5 equivalents of MI and to end with 0.74% impurity in the OND.base.cryst. When 5 equivalents of MI (but not 3 or 4) were employed, 10% Celite (added at the beginning of the reaction), was necessary to facilitate the filtration (OZ-417). The filtered cake was washed, dried and recrystallized in 30 volumes of methanol which was treated first with carbon. The celite already existed in the mixture, was also very helpful in the filtration during the carbo treatment.

An experiment was made to combine both reaction and crystallization into one step by adding methanol at the end of the reaction and crystallizing the product from water/methanol mixture with carbo (OZ-401). The result was higher yield of the OND.base.cryst (74% vs. 66% in AD-283) but respectively higher level of exo-methylene was obtained (3.64% vs. 0.74% in AD-283).

An attempt to synthesize OND.base directly from Me-CAR by Mannich reaction, using paraformaldehyde, dimethylamine hydrochloride and methylimidazole was made (EB-295, OZ-285). Only 6% OND.base was obtained while the major products were DMA-Me-CAR, 67% and exo-methylene 22% (HPLC area percent).

ONDANSETRON HYDROCHLORIDE DIHYDRATE (OND.HCl.$2H_2O$)



ondansetron base (OND–base)

ondansetron hydrochloride dihydrate[19] (OND·HCl·$2H_2O$)

There are two procedures in Glaxo patents for OND.HCl.$2H_2O$ synthesis :

a) [1b]OND-base in a hot mixture of IPA (5.4 volumes) and water (1 volume) was treated with concentrated HCl (1.05 equivalents) and after filtration of the hot mixture it was diluted with IPA (5 volumes) and stirred at r.t. (17 hours) and filtered at 2°C to obtain OND.HCl.$2H_2O$ which was crystallized in IPA/H_2O (1:1.7 v/w) to obtain 96% overall yield.

b) [24]OND.base in a mixture of IPA/H_2O/AcOH (8.9:2.1:0.5 v/w) was heated to 70°C, HCl (1.05 equivalents) was added and the solution was cooled to 5°C, filtered and washed with IPA (4 volumes) and dried to obtain 88% OND.HCl.$2H_2O$.

On repeating procedure 'a' we obtained only 83% yield with 0.4% exo-methylene by-product. Recrystallization from IPA/H_2O gave 83% yield with 0.12% exo-methylene by-product (EB-312). Procedure 'b' gave 83% yield with 0.23% exo-methylene by-product (EB-297). In both experiments the color of OND.HCl.$2H_2O$ was not satisfactory (color index more than 0.15, 10% w/v, 430nm).

In our effort to improve the quality of OND.HCl.$2H_2O$ we found that crystallization with water and carbo had a great effect on both the color and the by-product content. Among the carbo which we examined (SX-2, EB-341-B; CA-1, EB-341-C; CXV, AD-284-3; SX-1, EB-341-A) SX-1 gave the best performance. SX-1 dramatically lowered the exo-methylene levels and improved the color.

TABLE 2

PURIFICATION OF OND.HCl.$2H_2O$ FROM exo-METHYLENE BY-PRODUCT WITH SX-1 CARBO

ENTRY	REFERENCE OZ-	% SX-1 CARBO	AREA % OF EXO-METHYLENE (HPLC)			
			OND BASE	OND.HCl.$2H_2O$ CRUDE	OND.HCl.$2H_2O$ PURE	
					FIRST CRYSTAL.	SECOND CRYSTAL.
1	442	5	0.88	0.70	-	0.05
2	451	5	0.90	0.68	-	0.01
3	441	5	1.40	0.93	-	0.06
4	452	5	1.30	1.30	-	0.05
5	444	5	1.74	1.43	-	0.20
6	453	5	1.68	1.67	-	0.14
7	456-A	5	-	2.40	1.30	0.54
8	456-B	10	-	2.40	0.70	0.16
9	456-C	15	-	2.40	0.42	0.05
10	455	5	0.95	1.02	-	0.19
11	457	5	0.85	0.35*	-	0.01

*OND.HCl2H_2O crude was prepared with 5% SX-1.

The potency of SX-1 type of carbo (manufactured by NORIT) to purify OND.HCl.$2H_2O$ was examined by several experiments using various amounts of SX-1 for two or three times in a row. The results are summarized in Table 2. We can see that the more SX-1 employed in the crystallization process (5, 10 and 15%) the purer OND.HCl.$2H_2O$ obtained (entry 7, 8 and 9). We also see that usually there was no purification in the hydrochlorina- tion step (for instance entry 4 and 6 and to a lesser extent entry 1, 2 and 5, entry 3 is an exception) unless SX-1 was used. Thus when carbo was used in the hydrochlorination step in addition to the two successive carbo recrystallizations, a much better result was obtained (entry 11 vs. 10).

In all the experiments in Table 2, the color index met the limit we set (0.15) even in OND.HCl.$2H_2O$ that contained more than 0.1% exo-methylene.

The conclusions we came to were that we can use a combination of a number of crystallizations with various amounts of SX-1 carbo (5, 10 or 15%) depending upon the amount of exo-methylene in OND.base.cryst. For OND.base.cryst. with 0.74% exo-methylene by-product which was obtained in the reaction with 5 equivalents MI (see previous chapter), 3 times 5% (entry 11, table 2) or twice 10% SX-1 carbo (entry 8, table 2) seems to be enough for a highly pure OND.HCl.$2H_2O$. We examined these processes (3x5% and 2x10%) in laboratory batches starting from OND.base containing 1.02% exo-methylene (OZ-473) and we found that both ways gave a 100% pure OND.HCl.$2H_2O$ (table 3, entries 2, 3 for 3x5% and entries 4, 5 for 2x10%). However, the 2x10% carbo crystallization is a better choice because of the higher overall yield (62% vs. 55% in 3x5%). We also examined quantitatively the possibility of preparing OND.HCl.$2H_2O$ by first making OND.base cryst. in the combined manner (crystallization from the reaction mixture) followed by 3x10% preparation/crystallization sequence of OND.HCl.$2H_2O$. The results are summarized in Table 3, entries 6-9. The exo-methylene content in OND.base.cryst obtained in this manner was higher in comparison to the standard procedure (3.46% in OZ-477 or 3.64% in OZ-401 vs. 1.02% in OZ-473) and the following 3x10% carbo treatments in the preparation and crystallizations gave only 40% OND.HCl.$2H_2O$ of a borderline quality.

TABLE 3

ENTRY	REFERENCE OZ-	% SX-1 CARBO	HPLC AREA %				NOTES
			OND	exo-METHYLENE	DMA-Me-CAR	Me-CAR	
1	473	-	98.37	1.02	0.34	0.08	OND.BASE PREPARATION , S.M. FOR ENTRIES 2-5.
2	475-1	5	99.28	0.57	0.09	0.06	OND.HCl.$2H_2O$ PREPARATION WITH 5% CARBO.
3	475-2	5+5	100.00	-	-	-	TWO CONSEQUENTIAL CRYSTALLIZATIONS OF 475-1.
4	476-1	0+10	98.73	0.98	0.10	0.09	OND.HCl.$2H_2O$ PREPARATION WITHOUT CARBO + ONE 10% CARBO CRYSTALLIZATION.
5	476-2	10	100.00	-	-	-	CRYSTALLIZATION OF OND.HCl.$2H_2O$.
6	477	-	95.88	3.46	0.19	0.35	OND.BASE PREPARATION *, S.M. FOR ENTRY 7-9.
7	480-1	10	97.71	1.94	0.07	0.30	OND.HCl.$2H_2O$ PREPARATION WITH 10% CARBO
8	480-2	10	99.31	0.54	-	0.15	FIRST CRYSTALLIZATION OF OND.HCl.$2H_2O$
9	480-3	10	99.86	0.09	-	0.05	SECOND CRYSTALLIZATION OF OND.HCl.$2H_2O$

* OND.base preparation without separation of the crystallization from the reaction